and phosphatases, would have a dramatic effect on cell components.

Although the pattern of breakdown of many cell consituents is similar to that found during natural senescence but on a shorter time-scale, significant differences in comparative times of changes in membrane permeability and photosynthetic activity (Baldwin, Dodge & Harris, 1968; Drury & Park, 1968) indicate that the herbicidal activity is a fundamentally different process.

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The Biochemistry of Warfarin Resistance in the Rat

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Warfarin-resistant rats were first reported on farms in Scotland (Boyle, 1960) and have since appeared in Denmark (Lund, 1964) and in areas of Montgomeryshire and Shropshire (Drummond, 1966). The resistance is inherited as a single autosomal dominant gene, which is linked to coat colour (Greaves & Ayres, 1967, 1969).

Whereas in normal rats ingestion of warfarin results in the blood's becoming incoagulable, in resistant animals blood clotting times are only slightly prolonged. It is now generally accepted that the sole function of vitamin K in animals is to maintain the normal synthesis of clotting factors II, VII, IX and X; the effect of warfarin is to decrease the concentrations of these factors in the plasma. Warfarin has also been shown to inhibit the production of factor VII *in vitro* by rat liver slices (Pool & Borchgrevink, 1964; Lowenthal & Birnbaum, 1969).

The distribution of radioactivity in rats after the intraperitoneal administration of [4-^{14}C]warfarin has been followed (Link, Berg & Barker, 1965) and the metabolites in the urine identified (Barker, Hermodson & Link, 1969). In addition to unchanged warfarin, these comprise the 6-, 7-, 8- and 4′-hydroxywarfarin, 7-hydroxywarfarin glucuronide and an intramolecular cyclization product, 2-methyl-4-phenyl-3,4-dihydropyrano[3,2-*c*][1]-benzopyran-5-one.

The present comparisons of various aspects of warfarin metabolism in normal and warfarin-resistant rats were facilitated by the synthesis of the following radioactively labelled warfarin species: [3-^{14}C]warfarin (0.80mCi/mmol), [α-^{14}C]warfarin (4.0mCi/mmol), [*carbonyl*-^{14}C]warfarin (0.87mCi/mmol) and warfarin generally labelled with tritium in the aromatic moiety of the coumarin nucleus (5.4mCi/mmol). The total radioactivity in urine and faeces and the specific radioactivities of plasma, liver and kidneys of both types of rat were determined at intervals after single oral doses of warfarin varying from 0.2 to 100mg/kg. Extracts of urine, plasma and liver were subjected to t.l.c. and the metabolites were compared by radioautography and by the use of spray reagents. Quantitative results from resistant animals were comparable with normal values, although the latter varied considerably from animal to animal. No significant differences in the nature and relative amounts of warfarin metabolites in plasma, liver or urine could be found by the methods employed. These initial observations, which suggested that the rate and mode of warfarin detoxication after a single dose are the same in normal and resistant rats, confirmed those of Pool, O'Reilly, Schneiderman & Alexander (1968) and Hermodson, Suttie & Link (1969).

The LD_{50} for warfarin administered on a multiple-dose basis, however, is much lower than the acute LD_{50} (Krieger, 1949; Hayes & Gaines, 1950; Berg, 1964; Pyörälä, 1968), and it is on a multiple-dose basis that resistant rats are selected in the field (Drummond & Wilson, 1968). Three resistant and three normal rats were each given four daily oral doses (0.2mg/kg) of [α-^{14}C]warfarin. Samples of plasma, urine and faeces were collected 12h after each dose and assayed for radioactivity; 84h after the initial dose the average plasma concentration of radioactivity was 1.65% of a single dose for the normal and 1.49% for the resistant rats. Urinary excretion accounted for 27.3% of the radioactivity of the total dose in normal and 26.5% in resistant animals. However, whereas 13.5% of the total dose appeared in the faeces of normal rats, the corresponding value for the resistant group was 24.3%. These results do not completely exclude differences between the mechanisms of warfarin detoxication in the two types of rat. Each of the metabolites of warfarin in the urine accounts for at least 5% of the administered dose (Barker *et al.* 1969) and the present work shows the amount of these metabolites

in liver and plasma to be of this magnitude, although only 0.2% of a dose of [*carbonyl*-^{14}C]warfarin had appeared in the expired carbon dioxide of both normal and resistant rats after 24h.

The conversion of warfarin into 6-, 7- and 8-hydroxywarfarin by rat liver microsomal preparations (Ikeda, Ullrich & Staudinger, 1968) has been confirmed; comparison with a similar preparation from a resistant rat revealed no difference in the rate of formation of these metabolites. Comparisons by other workers of enzyme activities between the two strains of rat have also been reported (Taylor & Townsend, 1970).

Menadione is converted by rats into menaquinone-4 (Martius & Esser, 1958; Taggart & Matschiner, 1969). An analogous reaction could convert warfarin into a lipophilic metabolite that is the true antagonist of vitamin K at the receptor site, and such a conversion may be inefficient in resistant rats. In a large number of experiments the liver lipid from normal rats fed on both large and small, single and multiple, doses of labelled warfarin was chromatographed and the fractions were assayed for radioactivity. Less than 0.005% of the administered dose appeared in the fractions that would contain vitamins K_1 and K_2; some radioactivity, however, was associated with compounds less polar than warfarin, and a detailed examination of liver lipid from both normal and resistant rats is being carried out. A recent report (Matschiner, Amelotti, Bell & Knauer, 1969) suggests that the administration of warfarin does in fact affect vitamin K metabolism in normal rats.

Warfarin-resistant rats, rendered vitamin K-deficient, required some 20 times as much vitamin K_1 as normal rats to correct the deficiency (Hermodson *et al.* 1969). The most popular hypothesis on the mode of action of warfarin proposes that the anticoagulant competes directly with vitamin K for a receptor site. Hermodson *et al.* (1969) suggest that warfarin resistance might be explained in terms of a receptor site that, in resistant rats, has only a low affinity for warfarin and vitamin K.

Whatever the true mechanism of warfarin resistance, a study of these animals should be of considerable value in resolving the prevailing confusion about the mode of action of vitamin K and its antagonists.

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Kinetics and Metabolism of Chlorinated Insecticide Residues

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The environmental contamination by chemicals is generally identified by 'residue levels'. While this term has been defined by the FAO/WHO Joint Meeting for pesticide residues, it may have various meanings when applied to other environmental chemicals. Sometimes only parent compounds are involved, but sometimes biotic or abiotic conversion products also. When the term 'general residue levels' is used for the description of environmental quality, this includes different parameters, e.g. concentration of parent compounds, persistence, potential toxicity, biochemical and chemical transformation etc.

The complexity of environmental problems is also a reason for not correlating the term 'persistence' in a generalized way with special classes of chemical compounds. Although many cyclodiene insecticides are persistent in soil, water, plants and animals as the parent compounds, some breakdown products of easily hydrolysed or oxidized insecticides (so-called non-persistent ones) are more persistent than the cyclodiene insecticide β-dihydroheptachlor.